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Collagen Thermal Damage and Collagen Synthesis After Cutaneous Laser Resurfacing

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Background and Objective: **Objectively measure and compare postoperative collagen thermal damage and subsequent new collagen synthesis after cutaneous laser resurfacing using two carbon dioxide laser systems.**
Study Design/Materials and Methods: **We created 240 resurfacing wounds on eight piglets with scanned and short-pulsed lasers using the manufacturer's suggested settings. The wounds varied with respect to the number of laser passes and postoperative survival times. Samples were harvested for histological analysis.**
Results: **The scanned laser resulted in an average of 52% more collagen thermal damage on the day of surgery ($P < 0.0001$) and an average of 78% more thermal damage 3 days postoperative ($P < 0.0001$) than the short-pulsed laser. The amount of new collagen synthesis correlated with the amount of thermal damage, with the scanned laser wounds showing 44% greater new collagen synthesis than the short-pulsed laser wounds on postoperative day 7 ($P < 0.0001$) and 48% greater new collagen synthesis on postoperative day 14 ($P < 0.0001$).**
Conclusion: **Compared to the short-pulsed laser, the scanned laser results in a greater depth of collagen thermal damage with a correspondingly greater depth of new collagen synthesis after cutaneous resurfacing. Lasers Surg. Med. 23:66–71, 1998.**

Key words: **histology; pulse structure; animal model; morphometric analysis; comparison study**

INTRODUCTION

For almost 100 years, clinicians have traditionally used either dermabrasion or chemical peels to improve facial cutaneous disorders, including fine wrinkles caused by aging and solar exposure [1–3]. However, these methods have been criticized for their lack of control over the depth of injury to the skin when factors such as skin hydration, chemical concentration, and operator-skill variables are introduced. Complications such as hypopigmentation and scarring occur when the injury extends too deeply into the dermis. Therefore, the carbon dioxide (CO_2) laser has recently been adapted as a cutaneous resurfacing tool to gain more control over the depth of injury and to avoid these complications [4].

The goal of any cutaneous resurfacing technique is the removal of the superficial epidermis and papillary dermis while avoiding the reticular dermis. The organized collagen synthesis that oc-

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curs after the dermis is damaged is widely accepted to be responsible for the clinical improvement of fine wrinkling [1,2]. Histologically, resurfaced skin demonstrates a thicker dermis with more parallel bands of collagen and less elastosis and perivascular inflammation [3,5]. However, if the ablation extends too deeply into the reticular dermis, unwanted excessive scarring with disorganized collagen bands and lasting postoperative erythema occurs [6]. Current CO_2 laser technology allows for the destruction of superficial layers of skin and controlled induction of thermal damage to the underlying papillary dermis. Computerized, scanning delivery systems and shorter pulse durations allow for well-defined depth and breadth of thermal injury caused by the laser regardless of the patient skin variability. It has recently been shown that selective ablation of skin with minimal collateral thermal injury occurs when less than 5 J/cm^2 is delivered in under 1 millisecond to the surface [7]. Both the scanned and short-pulsed lasers were developed to satisfy this requirement by delivering high-powered laser energy to the skin in a short enough time frame to avoid collateral damage [8].

While both useful in improving fine wrinkles on the face, the scanned laser is reported to cause greater postoperative erythema than the short-pulsed laser [9]. The undesirability of this erythema has led many facial surgeons to prefer the short-pulsed laser, although the relative effectiveness between the two lasers is not well characterized.

This study is designed to compare the two laser systems with specific attention to the amount of collagen thermal damage and subsequent new collagen synthesis after cutaneous resurfacing.

MATERIALS AND METHODS

A porcine model for human skin [10] was used and institutional guidelines regarding animal experimentation were followed. Piglets were anesthetized with halothane during the laser treatment. Prior to laser resurfacing, the piglets were shaved and then cleansed with antibacterial soap and saline. Fifteen wounds were created on each side of the piglet using the scanned or short-pulsed laser systems. The scanned laser was a Sharplan Silktouch (Sharplan Lasers Inc., Allendale, NJ) with a 125 handpiece (200 micron spot size) coupled to a Sharplan 1060 continuous wave CO_2 laser (Sharplan Lasers Inc., Allendale, NJ) set to repeat pulses (approximately a 1.3 Hz repetition rate), 8 Watt, 0.2 second pulses. The short-pulsed laser was a TruPulse CO_2 laser (Tissue Technologies, Albuquerque, NM) set to single, 500 mJ pulses with a 5 Hz repetition rate. The TruPulse spot size was 3.0 × 3.0 millimeters.

Wounds of equal surface area were created with one, three, or five passes with each laser. Between each pass, the wounds were debrided with a saline-soaked gauze pad. The wounds were then covered with one of four dressings, including Vaseline (Cheseborough Ponds, Greenwich, CT), triple antibacterial ointment, vegetable oil, or Vigilon (Bard Urological, Covington, GA) wound dressing. The wounds were then covered with a transparent dressing (Opsite, Smith-Nephew, Memphis, TN) and wrapped with Coban Elastic Bandage (3M, St. Paul, MN) to prevent contamination of the wounds.

Each piglet had five of each wound type (2 lasers; 1, 3, or 5 passes) for a total of 30 wounds per piglet. Two piglets were sacrificed on the day of surgery, at 3 days post-surgery, at 7 days post-surgery, and at 14 days post-surgery. Thus, all histological data represents information obtained from two different animals.

The tissue was stained with Masson's Trichrome to evaluate the nature and depth of collagen damage and new collagen synthesis. This depth of collagen damage or synthesis was viewed at 200 power magnification with a Zeiss Axioplan microscope (Carl Zeiss, Thornwood, NY) and measured using Image Pro Plus for Windows 2.0 (Media Cybernetics, Silver Spring, MD). Statistical analysis was performed with Statview 4.0 (Abacus Concepts, Berkeley, CA).

RESULTS

No differences were seen among the different methods of postoperative care [9,10]. Figure 1 shows representative histology from the scanned and short-pulsed laser wounds harvested on the day of resurfacing (A, B) and postoperative day 7 (C, D). Histological analysis of the skin samples revealed statistically significant differences between the scanned and short-pulsed lasers with regards to thermal collagen damage and new collagen synthesis after resurfacing (Fig. 2). Each value represents the average of data collected from at least three skin samples. The statistical significance values of the differences between the scanned and short-pulsed lasers are included.

In wounds harvested the day of the resurfacing, one pass of the scanned laser produced 78 ± 6

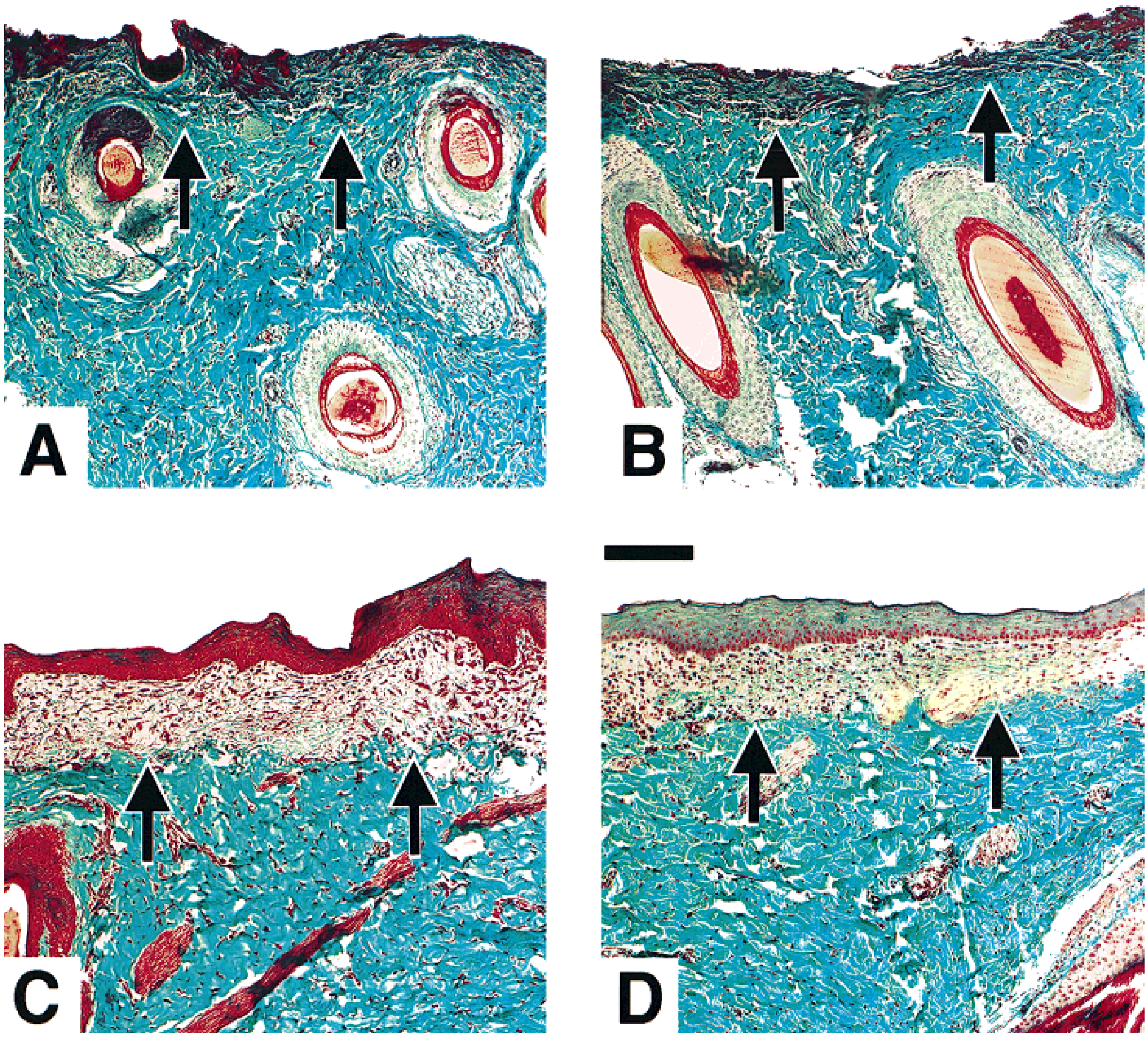

Fig. 1. Representative histology of tissue sections treated with five passes of the respective lasers and harvested on the day of resurfacing or postoperative day 7. Masson's Trichrome stain, all sections photographed at 50×. **A.** Scanned laser wound harvested on day of resurfacing. **B.** Short-pulsed laser wound harvested on day of resurfacing. **C.** Scanned laser wound harvested on postoperative day 7. **D.** Short-pulsed laser wound harvested on postoperative day 7. Arrows indicate lower extent of collagen thermal damage or collagen synthesis. Scale bar = 100 μm.

μ of thermal damage compared to 46 ± 2 μ of thermal damage produced by the short-pulsed laser ($P < 0.001$). Three passes of the scanned laser produced 77 ± 4 μ of thermal damage compared to 54 ± 6 μ of thermal damage produced by the short-pulsed laser ($P < 0.004$). Five passes of the scanned laser produced 80 ± 2 μ of thermal damage compared to 54 ± 11 μ of thermal damage produced by the short-pulsed laser ($P < 0.016$).

In wounds harvested 3 days after resurfacing, one pass of the scanned laser produced 96 ± 15 μ of thermal damage compared to 53 ± 11 μ of thermal damage produced by the short-pulsed laser ($P < 0.017$). Three passes of the scanned laser produced 91 ± 7 μ of thermal damage compared to 55 ± 9 μ of thermal damage produced by the short-pulsed laser ($P < 0.004$). Five passes of the scanned laser produced 102 ± 19 μ of thermal damage compared to 54 ± 3 μ of thermal damage produced by the short-pulsed laser ($P < 0.013$).

In wounds harvested 7 days after resurfacing, 108 ± 10 μ of new collagen was observed with one pass of the scanned laser compared to 77 ± 8 μ of new collagen was observed with one pass of the short-pulsed laser ($P < 0.030$). Wounds created with three passes of the scanned laser had 116 ± 12 μ of new collagen compared to 73 ± 4 μ of new collagen in wounds created with the short-

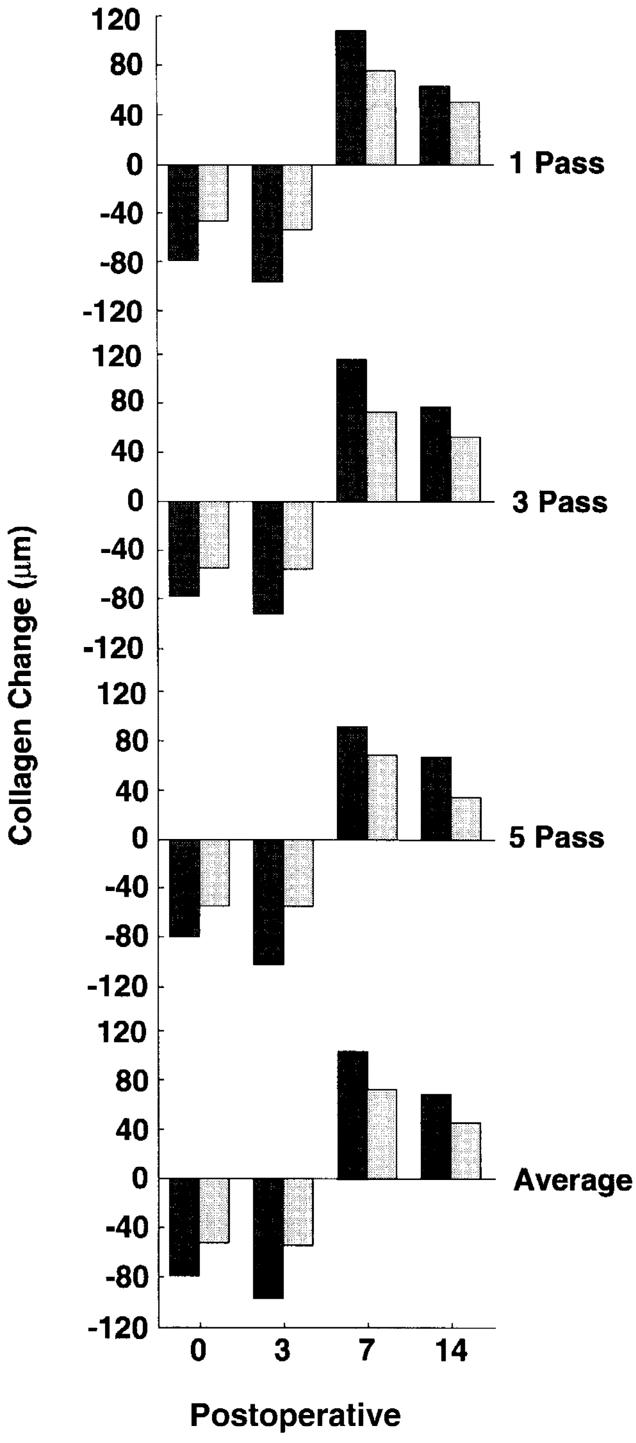

Fig. 2. Comparison of collagen thermal damge and new collagen synthesis after cutaneous resurfacing with the scanned and short-pulsed carbon dioxide (CO_2) lasers. Comparisons of collagen effect after one, three, and five passes are shown. In the comparison graph of data averages, each value is an average of nine skin samples without regard to number of passes with the CO_2 laser. (–) Values indicate thermal collagen damage; (+) values indicate new collagen synthesis. Dark-shaded columns indicate the scanned laser; light-shaded columns indicate the short-pulsed laser.

pulsed laser ($P < 0.001$). Wounds created with five passes of the scanned laser had 93 ± 8 μ of new collagen compared to 70 ± 5 μ of new collagen in wounds created with the short-pulsed laser ($P < 0.003$).

In wounds harvested 14 days after resurfacing, 64 ± 7 μ of new collagen was observed with one pass of the scanned laser compared to 51 ± 3 μ of new collagen observed with one pass of the short-pulsed laser ($P < 0.014$). Wounds created with three passes of the scanned laser had 78 ± 5 μ of new collagen compared to 53 ± 6 μ of new collagen in wounds created with the short-pulsed laser ($P < 0.001$). Wounds created with five passes of the scanned laser had 68 ± 14 μ of new collagen compared to 35 ± 3 μ of new collagen in wounds created with the short-pulsed laser ($P < 0.016$).

Overall, in piglets sacrificed 3 days after resurfacing, wounds created by the scanned laser revealed from 65% to 88% greater thermal collagen damage than short-pulsed laser wounds. Additionally, in piglets sacrificed 7 days after resurfacing, wounds created by the scanned laser revealed from 32% to 58% more new collagen synthesis than short-pulsed laser wounds.

DISCUSSION

CO_2 lasers can provide unique advantages to traditional methods in the battle against wrinkles, acne, and other benign conditions of the face. The precise control over depth of thermal damage translates into more consistent and reliable results with respect to efficacy and outcomes. The adjustability of laser settings also allows the physician to tailor the treatment to the patient and the situation, again enhancing the performance of this procedure. However, it has been difficult to compare and assess the performance and benefits of different lasers on the market today. Rarely will one physician use several different lasers for the treatment of the same condition and even more rarely will a patient allow treatment with two different lasers during the same visit.

Because a controlled comparison of thermal damage and subsequent dermal healing is difficult with human subjects, we used a porcine model to compare the collagen damage and synthesis affected by scanned and short-pulsed laser resurfacing. While comparisons between the thermal damage depth created by the scanned and short-pulsed lasers has been done in human skin [5,11,12], we believe that our in vivo experiments with piglets have clinical relevance. Porcine skin

TABLE 1. Average Depth of Collagen Effect*

Postoperative day	Scanned laser collagen effect (μm)	Short-pulsed laser collagen effect (μm)	*P* value
0	−78 ± 1	−52 ± 2	0.0001
3	−96 ± 5	−54 ± 2	0.0001
7	+105 ± 4	+73 ± 2	0.0001
14	+70 ± 3	+47 ± 3	0.0001

*(−)Values indicate thermal collagen damage; (+) values indicate new collagen synthesis.

has been shown to be very similar to human skin in such parameters as hair growth, thickness, and sweat glands [13,14]. Furthermore, parameters such as the roles of angiogenesis and inflammatory cells in collagen removal or synthesis can only be accounted for in an in vivo model. We note that these young piglets probably exhibit faster wound healing than an older, adult human patient. Still, the comparisons between the techniques are valid even if the absolute time durations are not.

When the skin samples were examined, we observed that the depth of thermal collagen damage peaks 3 days after initial skin resurfacing. Then, between postoperative days 3 and 7, the damaged collagen is cleared and a transition is made to collagen synthesis which continues on to postoperative day 14. Overall, the short-pulsed laser resulted in less thermal collagen damage and less new collagen synthesis than the scanned laser on all survival times.

Our data confirm that the number of passes with the laser does not appreciably affect the depth of thermal collagen damage induced in the skin [15]. Table 1 records the average depth of thermal damage and new collagen synthesis seen with both lasers. Each value is an average of nine skin samples without regard to number of passes with the respective CO_2 lasers. On the day of surgery, the scanned laser wounds showed 52% greater thermal collagen damage than the short-pulsed laser wounds. At postoperative day 3, when the amount of thermal damage peaks for both laser systems, the scanned laser wounds showed 78% greater thermal collagen damage than the short-pulsed laser wounds. At postoperative day 7, the scanned laser wounds had 44% greater new collagen synthesis than the short-pulsed laser wounds. Finally, at postoperative day 14, the scanned laser wounds had 48% greater new collagen synthesis than the short-pulsed laser wounds. All differences in values between the scanned and short-pulsed lasers are statistically significant.

Several reports confirm that new collagen synthesis after cutaneous resurfacing accounts for improvements of fine wrinkles on the face [1,2,5,11,12]. Given that the scanned laser works deeper in the dermis and affects a greater depth of new collagen synthesis, one might expect that the scanned laser would be more efficacious than the short-pulsed laser in removing deeper wrinkles. However, this advantage is not without cost as the scanned laser causes deeper collagen thermal damage and has been reported to cause greater postoperative erythema [9,16]. Future studies will need to be conducted to clarify the relative risks and benefits of using the scanned and short-pulsed lasers in specific clinical situations.

The results from this paper allow us to conclude that the scanned laser induces more collagen thermal damage upon cutaneous resurfacing and that there is more new collagen synthesized during the healing phase when compared to the short-pulsed laser.

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